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# Microbial water quality in clean water tanks following inspection and cleaning

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**Abstract:** Increased bacterial counts are often registered in drinking water leaving clean water tanks after the tanks have been emptied, inspected and cleaned by flushing. To investigate the reason for the increased bacterial concentrations and consequently limit it, samples from two clean water tanks before, during and after cleaning of the tanks were analysed. Bacteria were quantified, the dominating bacterial groups were identified and re-growth potential in the water was estimated. Bacterial counts did not exceed drinking water guideline values but ATP concentrations in the water were high right after start-up of the tanks, which may indicate that a substantial part of the bacteria in the drinking water leaving the tanks originated from the sand filter. This was supported by 16S DNA analyses.

**Keywords:** Drinking water, Bacteria, Sand filter, 16S DNA, ATP, Microbiology

## Introduction

After inspection and cleaning of clean water tanks, bacterial counts often increase for a period of days when the tanks are started up. The cause of the increased bacterial numbers is not known, therefore the water utility, VCS Denmark, and DTU Environment have conducted a joint project to investigate microbial growth in two clean water tanks at the main water work in Odense, Denmark. It is the desire of VCS Denmark to be able to distribute the drinking water to consumers immediately after the tanks have been cleaned by flushing with clean water at a medium pressure and without the use of chlorine or hydrogen peroxide and without preceding discard of large volumes of drinking water. The drinking water is ground water based and there is no physical biological barrier such as UV-filter or supplying of chlorine at the outlet of the clean water tanks.

The project aims to investigate why increased bacterial counts are often registered after start-up of clean water tanks despite the fact that VCS Denmark follows strict hygienic procedures. Since increased bacterial counts are often an indication of contamination or bacterial regrowth in drinking water systems the aim was to investigate whether the increased bacterial counts are due to intruding bacteria from the external environment or due to naturally present drinking water bacteria, not constituting a health risk. DNA analyses were applied to investigate the population dynamics in the system, including sand filters, sediment and water leaving the sand filters and clean water tanks. A best practice procedure for inspection, cleaning and following start-up will be developed based on the results.

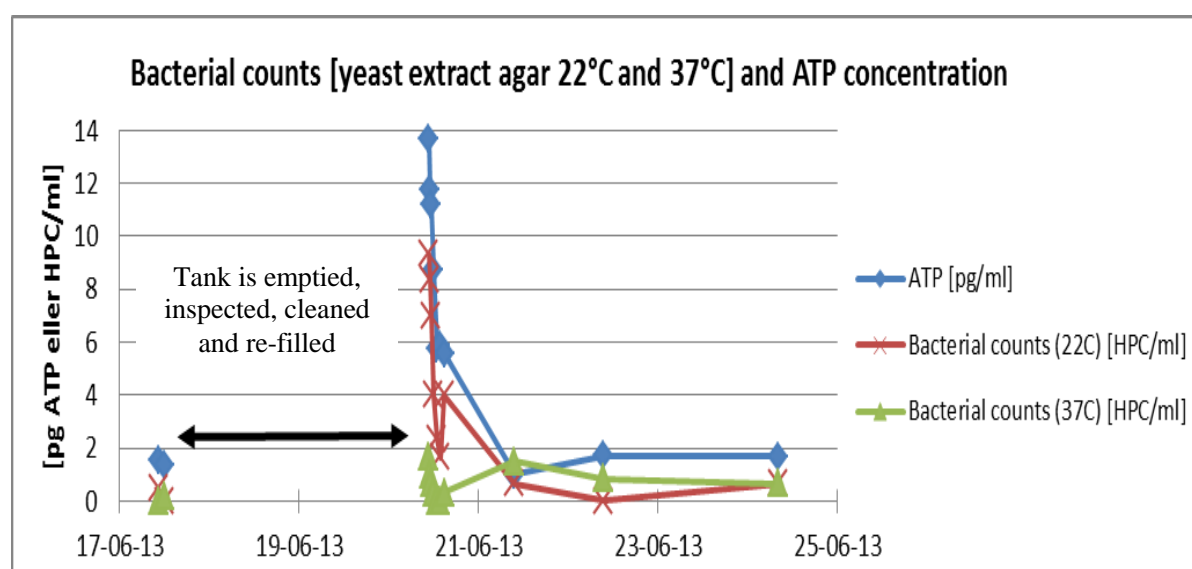
## Material and Methods

Sampling campaigns were carried out in two clean water tanks (April and June 2013) to investigate growth before, during and after cleaning of the tanks. Growth potential

and bacterial concentrations were quantified by ATP (Adenosin-Tri-Phosphate) measurements, Heterotrophic Plate Counts (HPC) on yeast extract agar (22° and 37°C), Colilert®, Enterolert® and Pseudalert®. Dominating bacterial groups were identified by cloning and 16S DNA sequencing of filtered water samples.

## Results and Conclusions

Bacterial counts [yeast extract agar, 22°C] did not exceed guideline values at any point during the investigations. Immediately after start-up, a maximum of 10 HPC/ml were measured in both tanks and after less than 24 hours the concentrations were below 1 HPC/ml in both tanks. The low concentrations were maintained the following days. Bacterial counts [yeast extract agar, 37°C] were below 2 HPC/ml at all times. To include bacteria not normally quantified by traditional methods, such as plating, we included ATP measurements of the water since ATP is an energy storage molecule present in all living bacteria.



**Figure 1. Concentration of ATP and bacterial counts [yeast extract agar, 22 and 37°C] in water leaving clean water tank 2 from the main water work at VCS Denmark.**

ATP concentrations in the water immediately after start-up were high compared to the concentration of culturable bacteria which may indicate that the bacteria from the sand filter e.g. nitrifying bacteria, which normally grow on substrates not present in the agar, are present in the water. Results from DNA analyses supported this conclusion but data are currently being analysed further in order to verify whether the bacteria present in the water after start-up are typical sand filter or groundwater bacteria or there could be an external source for the bacteria.